

AMENDMENTS TO THE CLAIMS

Please cancel claims 1-24 without prejudice or disclaimer of the subject matter set forth therein.

This listing of claims will replace all prior versions and listings of claims in the application:

Listing of claims:

1-24. (canceled)

Please add the following new claims:

25. (new) An isolated DNA molecule encoding a protein exhibiting alkaline liquefying α -amylase activity at a pH optimum of 8-9 and possessing an amino acid sequence which has been obtained by modifying an amino acid sequence described in SEQ ID NO:2 in a manner in which one amino acid is substituted, deleted, or inserted without changing enzymological properties of said amino acid sequence described in SEQ ID NO:2 and hydrolyzes 1,4- α -glucosidic linkages in starches, amylose, amylopectin, and degradation products thereof and in amylose forms: glucose (G1), maltose (G2), maltotriose (G3), maltotetrose (G4), maltopentose (G5) and maltohexose (G6) and does not hydrolyze pullulan.

26. (currently amended) The DNA molecule of claim 25, further comprising a nucleotide sequence for regulating expression of a DNA operatively linked to the DNA molecule.

27. (new) A recombinant DNA comprising the DNA molecule of claim 25.

28. (new) A recombinant DNA comprising the DNA molecule of claim 26.

29. (new) The DNA molecule of claim 25, wherein said encoded protein has an isoelectric point higher than 8.5 when measured by isoelectric focusing electrophoresis.

30. (new) The DNA molecule of claim 25, wherein said encoded protein:

is active in a pH range of 5.0 to 11.0, with an optimum pH in the range of 8.0 to 9.0;

is stable in a pH range of 5.0 to 10.5 and retains at least 50% of activity after treatment at 40°C for 30 minutes;

is active in a temperature range of 20°C to 80°C, with an optimum temperature in the range of 45°C to 55°C;

is stable at temperatures of 50°C or lower when treated for 30 minutes in a glycine-salt-sodium hydroxide buffer having pH

8.5;

has a molecular weight of 50,000+5000 when measured by sodium dodecyl sulfate polyacrylamide gel electrophoresis;

has an isoelectric point of approximately 9.2 when measured by isoelectric focusing electrophoresis;

is stable in the presence of K⁺, Na⁺, Ca²⁺, Mg²⁺, Mn²⁺, Ba²⁺, Fe²⁺, Fe³⁺, or Al³⁺; and

is substantially free of inhibition by surfactants selected from the group consisting of sodium linear alkylbenzene sulfonates, sodium alkylsulfonate esters, sodium polyoxyethylene alkylsulfate esters, sodium alkylsulfonates, soaps and polyoxyethylene alkyl ethers.

31. (new) The DNA molecule of claim 25, which encodes a protein exhibiting alkaline liquefying α -amylase activity at a pH optimum of 8-9, comprising at least one nucleotide sequence selected from the group consisting of SEQ ID NO: 10, SEQ ID NO: 7, SEQ ID NO: 3, SEQ ID NO: 6 and SEQ ID NO: 9.

32. (currently amended) The DNA molecule of claim 25, which encodes a protein exhibiting alkaline liquefying α -amylase activity at a pH optimum of 8-9 comprising at least one nucleotide sequence that is the reverse complement of a sequence selected from the group consisting of SEQ ID NO: 8, SEQ ID NO:

3, SEQ ID NO: 4 and SEQ ID NO: 11.

33. (new) The DNA molecule of claim 25, which encodes a protein exhibiting alkaline liquefying α -amylase activity at a pH optimum of 8-9 comprising at least one nucleotide sequence selected from the group consisting of SEQ ID NO: 10, SEQ ID NO: 7, SEQ ID NO: 3, SEQ ID NO: 6 and SEQ ID NO: 9, and also comprising at least one nucleotide sequence that is the reverse complement of a sequence selected from the group consisting of SEQ ID NO: 8, SEQ ID NO: 3, SEQ ID NO: 4 and SEQ ID NO: 11.

34. (new) An isolated DNA molecule encoding a protein exhibiting alkaline α -amylase activity at a pH optimum of 8-9 produced by a polymerase chain reaction using at least one of the primers selected from the group of SEQ ID NO: 10, SEQ ID NO: 7, SEQ ID NO: 3, SEQ ID NO: 6, SEQ ID NO: 9, SEQ ID NO: 8, SEQ ID NO: 4, and SEQ ID NO: 11 wherein said isolated DNA molecule is isolated from a member selected from the group consisting of *Bacillus* sp. A-40-2, *Bacillus* sp. NRRL B-3881, *Streptomyces* sp. KSM-9, *Bacillus* sp. H-167, *Bacillus* alkalothermophilus A3-8, and *Natronococcus* sp. Ah-36.

35. (new) The isolated DNA molecule of claim 34, wherein the DNA encodes a protein that is stable in a pH range of 5.0 to

10.5 and retains at least 50% of activity after treatment at 40°C for 30 minutes.

36. (new) The isolated DNA molecule of claim 34, wherein the DNA encodes a protein that is active in a temperature range of 20°C to 80°C, with an optimum temperature in the range of 45°C to 55°C.

37. (new) The isolated DNA molecule of claim 34, wherein the DNA encodes a protein that is stable at temperatures of 50°C or lower when treated for 30 minutes in a glycine-salt-sodium hydroxide buffer having pH 8.5.

38. (new) The isolated DNA molecule of claim 34, wherein the DNA encodes a protein that has a molecular weight of $50,000 \pm 5000$ when measured by sodium dodecyl sulfate polyacrylamide gel electrophoresis.

39. (new) The isolated DNA molecule of claim 34, wherein the DNA encodes a protein that has an isoelectric point of approximately 9.2 when measured by isoelectric focusing electrophoresis.

40. (new) The isolated DNA molecule of claim 34, wherein the DNA encodes a protein that is stable in the presence of K^+ , Na^+ , Ca^{2+} , Mg^{2+} , Mn^{2+} , Ba^{2+} , Fe^{2+} , Fe^{3+} , or Al^{3+} .

41. (new) The isolated DNA molecule of claim 34, wherein the DNA encodes a protein that is substantially free of inhibition by surfactants selected from the group consisting of sodium linear alkylbenzene sulfonates, sodium alkylsulfonate esters, sodium polyoxyethylene alkylsulfate esters, sodium alkylsulfonates, soaps and polyoxyethylene alkyl ethers.

42. (new) The isolated DNA molecule of claim 34, wherein the DNA encodes a protein that hydrolyzes 1,4- α -glucosidic linkages in starches, amylose, amylopectin, and degradation products thereof and in amylose forms: glucose (G1), maltose (G2), maltotriose (G3), maltotetraose (G4), maltopentose (G5) and maltohexose (G6) and does not hydrolyze pullulan.

43. (new) The isolated DNA molecule of claim 34, wherein the DNA encodes a protein that has an isoelectric point higher than 8.5 when measured by isoelectric focusing electrophoresis.